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MECHANISM OF ADAPTATION TO A THREONINE-DEFICIENT DIET, PART V

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(6) MECHANISM OF ADAPTATION TO A THREONINE-DEFICIENT DIET, V. Effect of quantity of sucrose and cellulose on the severity of threonine deficiency

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In a previous report (Arata, 1961), the suggestion was made that the severity of the fatty liver syndrome induced by a threonine deficiency was independent of dietary fat. When the fat level of a threonine-deficient diet was increased from 5% to 30%, the fatty liver syndrome was not worsened. In the act of increasing the dietary fat in this diet, the level of CHO was decreased. This stimulated the suggestion that the level of CHO, rather than the level of fat in the diet, was the causative agent in the production of fatty livers.

The experiment reported here represented an attempt to measure the relationship between the quantity of sucrose in the diet and the severity of the liver fat syndrome. Alphacel was used to replace a portion of sucrose in the diet, thus keeping other dietary constituents constant.

EXPERIMENTAL

Forty male weanling rats of the Sprague-Dawley strain were divided into 4 groups of 10 each. The rats were offered their respective diets ad libitum (Table I). Groups I and II were fed the standard 9% casein diet, except that tryptophan was not supplemented to either group. This amino acid was withdrawn from all 4 diets in an effort to remove the growth differential which usually exists between the threonine-deficient and threonine-adequate groups. Previous work indicated that the double amino acid deficiency does not influence the appearance of fatty livers in the threonine-deficient group (Arata, et al., 1954)

In groups III and IV, the same basic diet was used except that one-half of the sucrose was replaced with alphacel. All rats were allowed food and H_2O ad libitum for a period of two weeks. During the experimental period, data were collected on weight gain, food and water intake, and fecal output of the rats on the alphacel rations. On the 14th day the rats were sacrificed, and the livers were rapidly removed, weighed and homogenized in distilled water. Blood samples were collected from 4 representative rats in each group for serum protein analysis by paper electrophoresis.

TABLE I

Composition of Diets

Group	I	II	III	IV
Sucrose	81. 30	80. 94	40. 65	40. 47
Casein	09. 00	09. 00	09. 00	9. 00
D. L. Methionine	0. 30	0. 30	0. 30	0. 30
Tryptophan	------	------	------	------
Corn Oil *	5. 00	5. 00	5. 00	5. 00
Salts W. *	4. 00	4. 00	4. 00	4. 00
Vitamin Mix *	0. 25	0. 25	0. 25	0. 25
Choline	0. 15	0. 15	0. 15	0. 15
Alphacel	------	------	40. 65	40. 47
Threonine	------	0. 36	------	0. 36
Total %	100	100	100	100

* Composition given in previous research reports (Arata, 1960).

Liver homogenates were dried for 12 hours at 90° C and then ground, and 1 g samples were analyzed for fat by continuous fat extraction. The fat-free residues were stored for nitrogen determinations.

RESULTS

Data from analysis of livers from rats in all groups are compiled in Table II. A significant difference in liver fat levels was again observed between groups I and II. However, when one-half of the sucrose in these diets is replaced with alphacel (groups III and IV), liver fat levels are significantly ($P<0.01$) reduced in both the control groups (I vs. III) and the threonine-deficient groups (II vs. IV). Moreover, the severity of the fatty liver syndrome in rats fed a threonine-deficient diet containing alphacel is markedly reduced. Livers from threonine-deficient rats fed alphacel had liver fat levels only slightly deviated from their controls ($P>0.05$). No significant differences were observed in liver weights between any of the four groups.

Weight and food records are presented in Table III. No significant differences were recorded in any of the measurements between groups I and II or between groups III and IV. However, highly significant differences were observed when group I (no threonine, no alphacel) was compared with group III (no threonine, + alphacel) and when group II (+threonine, no alphacel) was compared with group IV (+ threonine, + alphacel).

Regardless of whether or not supplemental threonine was or was not present in the ration, the substitution of alphacel for one-half of the sucrose caused a significant increase ($P<0.01$) in weight gain, food intake, fecal output, and water consumption (Table III).

In the alphacel groups (III and IV), growth was stimulated over 150% compared with the animals fed a diet free of alphacel. In like manner, food intake was increased almost 200%, fecal output increased 15-fold, and H_2O intake increased by 200%.

DISCUSSION

A ration containing 40% sucrose and 5% fat as the sole source of calories for the young albino rat represents a suboptimal supply. In this experiment, the animals in groups III and IV (in which calories were essentially reduced in half)

TABLE II

Liver Data From Rats Fed 9% Casein Diets

	Sucrose		Sucrose + Alphacel	
	Group I (- threonine)	Group II (+ threonine)	Group III (- threonine)	Group IV (+ threonine)
Liver Fat (% dry wt.)	20.8±1.0*	10.2±0.5*	8.4±0.3*	7.1±0.5*
Liver Wts. (g)	4.68±0.20	4.81±0.28	4.98±0.24	5.22±0.24
Liver Moisture (%)	65.0±2.2	70.0±0.2	71.0±0.3	70.0±0.2

* Standard error of the mean.

TABLE III

Weight and Food Records for
Rats Fed a 9% Casein Diet

	Sucrose		Sucrose + Alphacel	
	Group I (-threonine)	Group II (+ threonine)	Group III (- threonine)	Group IV (+ threonine)
Wt. gain (g/week)	19±1*	23±2*	31±2*	35±2*
Final wt. (g)	78±2	85±3	103±3	110±3
Food Intake (g/week)	55±1	57±1	98±3	101±3
Food efficiency Ratio Wt. gain/g food	0.35	0.39	0.31	0.34
Fecal Output g/day	0.5	0.5	6.4	6.5
Water Intake ml/day	12	12	24	24

* Standard error of the mean.

automatically doubled their food intake, ostensibly to satisfy their caloric requirement. This need for calories must have been critical because of the tremendous volume of food and water consumed by the animals in groups III and IV. The distension in the gastrointestinal tract suggested that a certain amount of discomfort must have been experienced by these animals.

The failure of severe fatty livers to appear in threonine-deficient rats fed alphacel (group III, Table II) was not, therefore, a reflection of a decreased consumption of carbohydrates; but rather it reflected an increased consumption of protein. As these animals doubled their food intake in order to satisfy their calorie requirements, they also doubled their consumption of protein. As a result, the rats in groups III and IV were essentially consuming what amounted to an 18% casein diet. With the intake of casein this high, threonine is no longer as limiting as it is on a 9% casein diet.

This problem -- of how to adjust the diet in order to keep the total number of calories constant, but to vary the source of these calories from carbohydrate to fat, and yet to keep protein intake constant -- is currently under study. We have not yet thought of a method of accomplishing this directly. Two indirect methods have been devised; one is currently under study. The two methods suggested are:

1) To calculate the number of calories provided in the standard 9% casein diet. Dietary constituents would be adjusted in such a manner that in one diet 90% of those calories are provided by fat and 10% by sucrose. In subsequent diets, various percentages of the calories will come from fat and from sucrose. The weight differences will then be made up with alphacel, but all diets will be iso-caloric with the standard diet. In this situation, we should not run aground on the problem of marked differences in food intake.

2) To pair-feed the low carbohydrate group of rats with the rats fed the standard diet. We recognize the danger here of observing effects of semi-starvation, but the results of this experiment, with the one described above, may be informative.

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